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著者	Fukushi Tsukasa
journal or publication title	The bulletin of the Marine Biological Station of Asamushi, Tohoku University
volume	10
number	1
page range	57-63
year	1960-03-25
URL	http://hdl.handle.net/10097/00131093

BULLETIN OF THE MARINE BIOLOGICAL STATION OF ASAMUSHI,
VOL. X, NO. 1, 1960

THE EXTERNAL FEATURES OF THE DEVELOPMENT OF THE SEA URCHIN, *GLYPTOCIDARIS CRENULARIS* A. AGASSIZ[1)]

By

TSUKASA FUKUSHI
福士 尹
Marine Biological Station of Asamushi, Aomori Prefecture, Japan

The family *Phymosomatidae* is represented in recent seas only by a single genus and species, *Glyptocidaris crenularis*. This species is light cherry-brown in color, with long spines and the bare test measures 80 mm in maximum diameter. This species is known only from Northern Japan, ranging from Hakodate to Mutsu Bay on the east coast and to about Sado Island on the west coast (Mortensen 1935).

At Asamushi it is found on a hard sea bottom covered with thin mud in a depth of about 30 meters. The breeding season of *Glyptocidaris crenularis* at Asamushi is from the middle of February to the middle of April. The mature egg is spherical in shape, measuring approximately 125 microns in diameter, and is surrounded with a jelly coat. The polar bodies are often observable attaching on the surface of the egg. The egg is transparent and slightly salmon red in color. The eggs and sperms used for the observations on the development were obtained by the KCl method. No harmful effect was recognized in comparison with the development of the eggs taken out directly from the ovary. A time table of cleavage and development to the maximum pluteus without special feeding at 10°C are given below. The time is recorded from the moment of insemination.

Stage	Time	
Formation of fertilization cone		1 min.
Completion of fertilization membrane		5 min.
Disappearance of fertilization cone		10 min.
Monaster stage		45 min.
Streak stage	1 hr.	10 min.
Wrinkle stage	1 hr.	50 min.
Amphiaster stage	2 hrs.	25 min.
1st cleavage	3 hrs.	10 min.
2nd cleavage	4 hrs.	50 min.
3rd cleavage	6 hrs.	
4th cleavage	7 hrs.	40 min.
5th cleavage	9 hrs.	
6th cleavage	10 hrs.	30 min.
7th cleavage	12 hrs.	

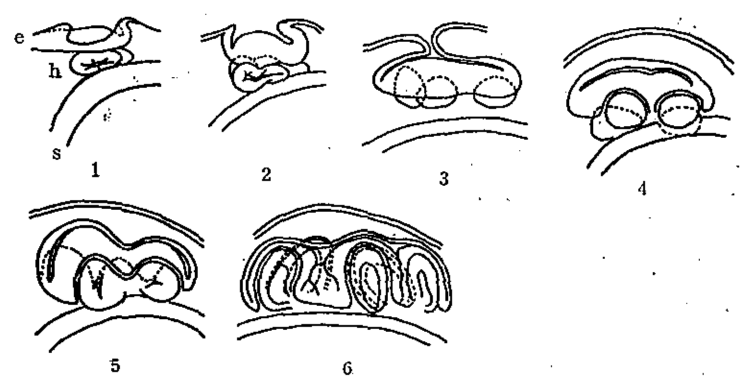
1) Contributions from the Marine Biological Station of Asamushi, Aomori Ken, No. 266.

Formation of cilia	21 hrs.
Hatching of blastula	27 hrs.
Formation of primary mesenchyme cells	36 hrs.
Aggregation of primary mesenchyme cells into two masses	49 hrs.
Skeleton begins	52 hrs.
Invagination begins	53 hrs.
Gut reaches animal pole	67 hrs.
Post oral rod begins	78 hrs.
Tripartition of gut begins	100 hrs.
Formation of left and right coelomic vesicles	120 hrs.
Formation of mouth	126 hrs.
Pluteus, maximum without special feeding	150 hrs.

The fertilization cone of the *Glyptocidaris* egg is relatively easy to observe and is as high as approximately 5 microns. It has no correlation with the position of the polar bodies, which are elevated attaching to the fertilization membrane. About 21 hours after fertilization the cilia of the surface of the blastula begin to beat. The thickness of the cell layer of the vegetal side becomes thick in comparison with that of the animal side, on which long cilia are observable at the time of hatching of the blastula. In the gastrula stage branches of the triradiate spicules formed in the centres of the mesenchymal masses elongate and become the antero-lateral, ventral transverse and body rod, respectively. The latticed rod of post oral arm arises as the parallel branches from the bases of three branches of initial spicules. The recurrent rod running back towards the posterior of the larva parallel to the body rod is given off as a posterior branch of the antero-lateral rod, and later connects with the body rod. The left and right recurrent rods meet each other at the posterior pole. About 90 hours after fertilization there is a ring of the secondary mesenchyme cells at the apex of the gut, and this later divides into two pouches to give rise to the left and right coelomic vesicles. The pluteus grows for six days after fertilization without special feeding, and thereafter, it does not show any development in this condition except for a slight elongation of the arms already formed.

For rearing the plutei until the completion of metamorphosis a glass vessel of 300 ml and 7 cm in depth was used. Fresh sea water taken from the sea was changed every day. The pluteus feeds mainly on *Peridinium* living in fresh sea water. The culture was carried out at the room temperature of 10°C to 25°C. The plutei begin to feed five days after fertilization in the room temperature and six days after in the thermostat regulated at 10°C. By the eighth day a pair of the postero-dorsal (pdr) and the pre-oral rods appear almost simultaneously as three cornered bodies. The former is situated at the lateral sides of the larva in the concavity between the oral lobe and post oral arms, and the latter, which is called the dorsal arch (da), appears on the dorsal side of the oesophagus. The pore canal (pc) is stretching from the left coelomic vesicle (lcv) towards the median line of the

dorsal side. The coelomic vesicle on each side becomes stretched to the posterior and divided into anterior and posterior portions at the side of the constriction between the oesophagus and stomach. By the tenth day a thick-walled outgrowth begins to appear at the posterior end of the left anterior coelomic vesicle. This is the hydrocoele (h), the rudiment of the adult water-vascular system (MacBride 1903, 1914b; von Ubisch 1913). This lies just beneath the ectoderm on the left side in the concavity between the postero-dorsal and post oral arms. The anterior end of the left posterior coelomic vesicle is closely attaching to the hydrocoele. Although it is difficult to confirm the dorsal pore in the *Glyptocidaris* larva, the left anterior coelomic vesicle seems to communicate with the exterior at the left side of the mid-dorsal line through the pore canal before the formation of the hydrocoele. The postero-dorsal rod is fenestrated. At this stage the length of the larva measures approximately 580 microns from the posterior pole to the tips of post oral arm. By the 17th day the ectoderm just above the hydrocoele becomes thick and invaginates forming the amniotic invagination (Fig. 1). Simultaneously the hydrocoele itself gives rise to the five lobes in the external feature. By the end of the third week the amniotic invagination becomes converted into a closed amniotic cavity (Fig. 4). Five processes grow inwards at the floor of the



Figs. 1–6. Amniotic invagination and the development of echinus rudiment. Figs. 1–3, during the third week after fertilization. A part of the ectoderm in contact with the hydrocoele becomes thick and invaginating. The hydrocoele is divided into five lobes. The openning of the amniotic invagination becomes gradually closed. Figs. 4–6, during the fourth week after fertilization. The invaginated ectoderm is cut off from the epithelium to form the amniotic cavity. The primary tube feet develop. e, ectoderm ; h, hydrocoele ; s, stomach.

amniotic cavity corresponding to the five lobes of the hydrocoele, which is closely attaching to it. Four pairs of arms grow longer so that their tips extend to the same extent from the body. There appears the postero-lateral processes (plp) with a strong ciliated band on each side of the posterior pole. They are supported by the posterior transverse rods (ptr). Although the dorsal pore is recognizable, the end of the pore canal is situated at the mid-dorsal line shifting from its original position on the left. A small ectodermal invagination appears inside the loop of the ciliary band on the right side. Later, in the invagination a pair of spines is formed. This invagination agrees with the "spine invagination" (sp) already reported by Runnström (1918) and Ohshima (1922) in the larva of *Psammechinus miliaris*. During the fourth week the echinus rudiment grows in size, and the lobes of the hydrocoele begin to appear as tube feet (Fig. 6) projecting into the amniotic cavity. The dorsal side of the stomach becomes covered by the networks of the spicules (ns) which grow from the dorsal arch and the recurrent rods. The ciliated bands between the antero-lateral and postero-dorsal arms, and post oral arms become conspicuous to grow up to the vibratile lobes (vl) and bands (vb). By the fifth week the tube feet begin to move in the amniotic cavity. Sometimes the amniotic veil gets torn and the tube feet cling to the bottom of the glass container with their suckers. The bases of the tube feet are covered with the adult spines (is). On the surface of the body the spines (es) become also formed. At first they appear at the posterior pole and between the post oral and postero-dorsal arms on the right side. The former is a single spine and the latters a pair of spines formed in the spine invagination cavity mentioned above. One or two days later, a pair of spines at the dorsal side of the oesophagus, and a single spine at the dorsal side of the stomach are formed. Simultaneously, a pedicellaria (p) and a pair of spines begin to appear at the right dorsal side. At the right side of the anal surface a single spine is formed. In *Glyptocidaris* the external spines and the spines formed in the amniotic cavity are similarly pointed, while the former is square-topped and the latter pointed in other species, for example *Psammechinus* (MacBride 1914a). The stomach (s) is pushed away to the right side because of the conspicuous enlargement of the echinus rudiment.

Now, the larva set up all of the indispensable accessories to metamorphose into a young sea urchin. The total size of the larva measures approximately 1.15 mm. By the first of the sixth week the earliest metamorphosing larva was found. The process of the metamorphosis is frequently observed soon after the time when the plutei of this stage were transferred to the dish with fresh sea water, and come to an end within one to two hours. The larva is situated on the left side down and clings to the bottom of the culture dish with the primary tube feet. The epithelial skin of the arms synchronously disappears from the tips leaving the supporting rods to project as naked spines. The projecting spines become broken off before long. The oral lobe shrinks and the mouth and oesophagus disappear. The anus also closes, and the stomach is isolated from the exterior until the new mouth and anus is opened. In the abactinal view of the imago soon after metamorphosis there are seen nine external spines (es) supported by calcareous plates, regularly arranged 30 spines (is), a pedicellaria (p) and five azygous tube feet (pt). At the central portion of the actinal surface nothing is found except a pentagon with five-radiate streaks around it. One or two weeks later, the tips of the incipient teeth appear in the centre of the pentagon and move to feed. Paired tube feet (st) grow first at the base of the azygous tube feet. It is different in individuals whether the first paired tube feet are already formed before metamorphosis. When they are absent by the time immediately after metamorphosis, they are formed within one or two days later at the normal position.

The writer wishes to thank Professor Isao Motomura for his constant guidance during the course of the present work. Thanks are also due to Dr. Eturô Hirai for his valuable advice and encouragement.

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EXPLANATION OF PLATES

LIST OF ABBREVIATIONS

a	anus
al	antero-lateral arm
br	body rod
da	dorsal arch
es	spine formed on the body surface of the larva
h	hydrocoele
is	spine formed in the amniotic cavity
lcv	left coelomic vesicle
m	mouth
ns	network of the spicule
o	oesophagus
p	pedicellaria
pc	pore canal
pda	postero-dorsal arm
pdr	postero-dorsal rod
plp	postero-lateral process
poa	post oral arm
pra	pre-oral arm
pt	primary azygous tube foot
ptr	posterior transverse rod
rcv	right coelomic vesicle
rr	recurrent rod
rs	rudiment of the spine
s	stomach
si	spine invagination
st	secondary paired tube foot
vb	vibratile band
vl	vibratile lobe

PLATE II

Fig. 1. Larva, eight days old, viewed from the abanal surface. ×83. Rudiments of the postero-dorsal and pre-oral rods appear.

Fig. 2. Larva, 14 days old, viewed from the abanal surface. ×84. Hydrocoele is already formed.

Fig. 3. Larva, 16 days old, viewed from the abanal surface. ×88. Pre-oral arms elongate. Amniotic invagination will occurs before long.

Fig. 4. Larva, 28 days old, viewed from the abanal surface. ×65. Primary tube feet develop. Spine invagination is shown. Networks of the spicules develop on the stomach.

PLATE III

Fig. 1. Larva, 36 days old just about to metamorphose, viewed from the abanal surface. ×61.

Fig. 2. The same specimen shown in Fig. 1, viewed from the anal surface. ×61.

Fig. 3. Abactinal view of a just metamorphosed imago. ×54.

Fig. 4. Actinal view of a imago four days old after metamorphosis. ×50. The mouth is not still opened.

PLATE IV

All figures magnified about ×128.

Fig. 1. Immature egg.

Fig. 2. Mature, unfertilized egg.

Fig. 3. The same with Fig. 2, showing the jelly layer by the aid of Chinese Black.

Fig. 4. Fertilized egg.

Fig. 5. Wrinkle stage.

Fig. 6. 2–cell stage.

Fig. 7. 4–cell stage.

Fig. 8. 8–cell stage.

Fig. 9. 16–cell stage. Polar view.

Fig. 10. 16–cell stage. Side view.

Fig. 11. 32–cell stage. Polar view.

Fig. 12. 32–cell stage. Side view.

Fig. 13. Blastula, early.

Fig. 14. Blastula, advanced.

Fig. 15. Hatching of blastula.

Fig. 16. Mesenchymal blastula.

Fig. 17. Gastrula. Side view.

Fig. 18. Gastrula. Polar view.

Fig. 19. Early pluteus. Abanal view.

Fig. 20. Early pluteus. Side view.

Fig. 21. Pluteus. Abanal view.

PLATE V

All figures magnified about × 43.

Fig. 1. Larva, eight days old, viewed from abanal surface.

Fig. 2. Larva, 10 days old, viewed from abanal surface.

Fig. 3. Larva, 16 days old, viewed from abanal surface.

Fig. 4. Larva, 28 days old, viewed from abanal surface.

Fig. 5. Larva, 36 days old, viewed from abanal surface.

Fig. 6. Larva, 36 days old, viewed from anal surface. Just before metamorphosis.

Fig. 7. During metamorphosis.

PLATE VI

All figures magnified about ×43, except Fig. 7, about ×90.

Fig. 1. During metamorphosis.

Fig. 2. During metamorphosis.

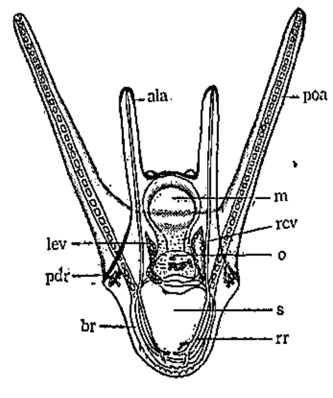
Fig. 3. Imago, just metamorphosed. Abactinal view.

Fig. 4. Imago, just metamorphosed. Actinal view.

Fig. 5. Imago, just metamorphosed. Side view.

Fig. 6. Imago, one week old after metamorphosis.

Fig. 7. Larva, 28 days old, viewed from left side, showing the five rudiments of the primary tube feet.



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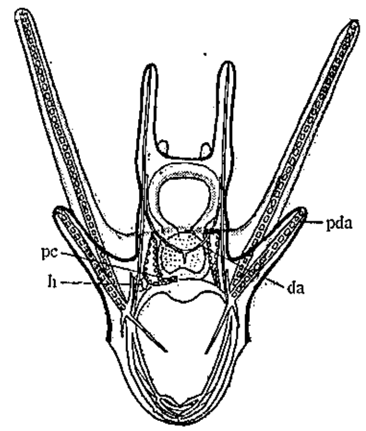

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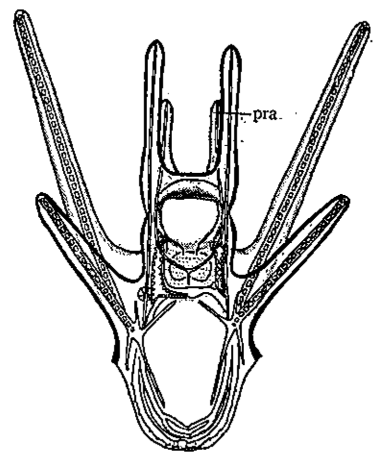

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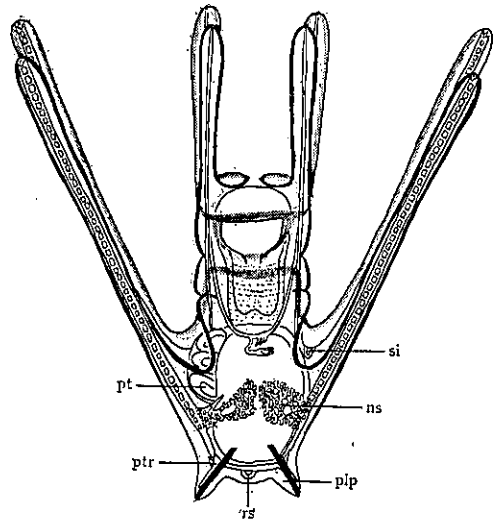

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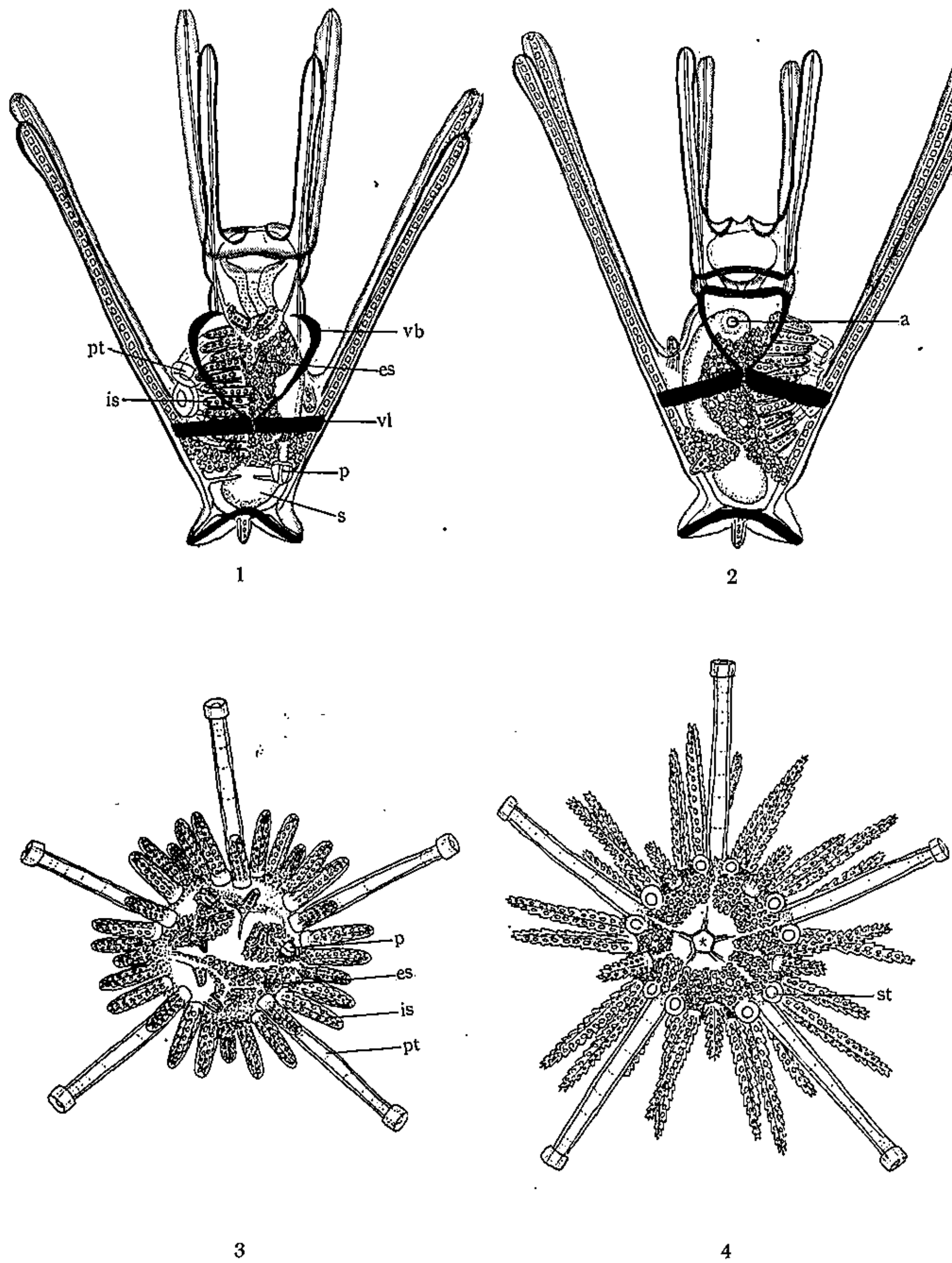

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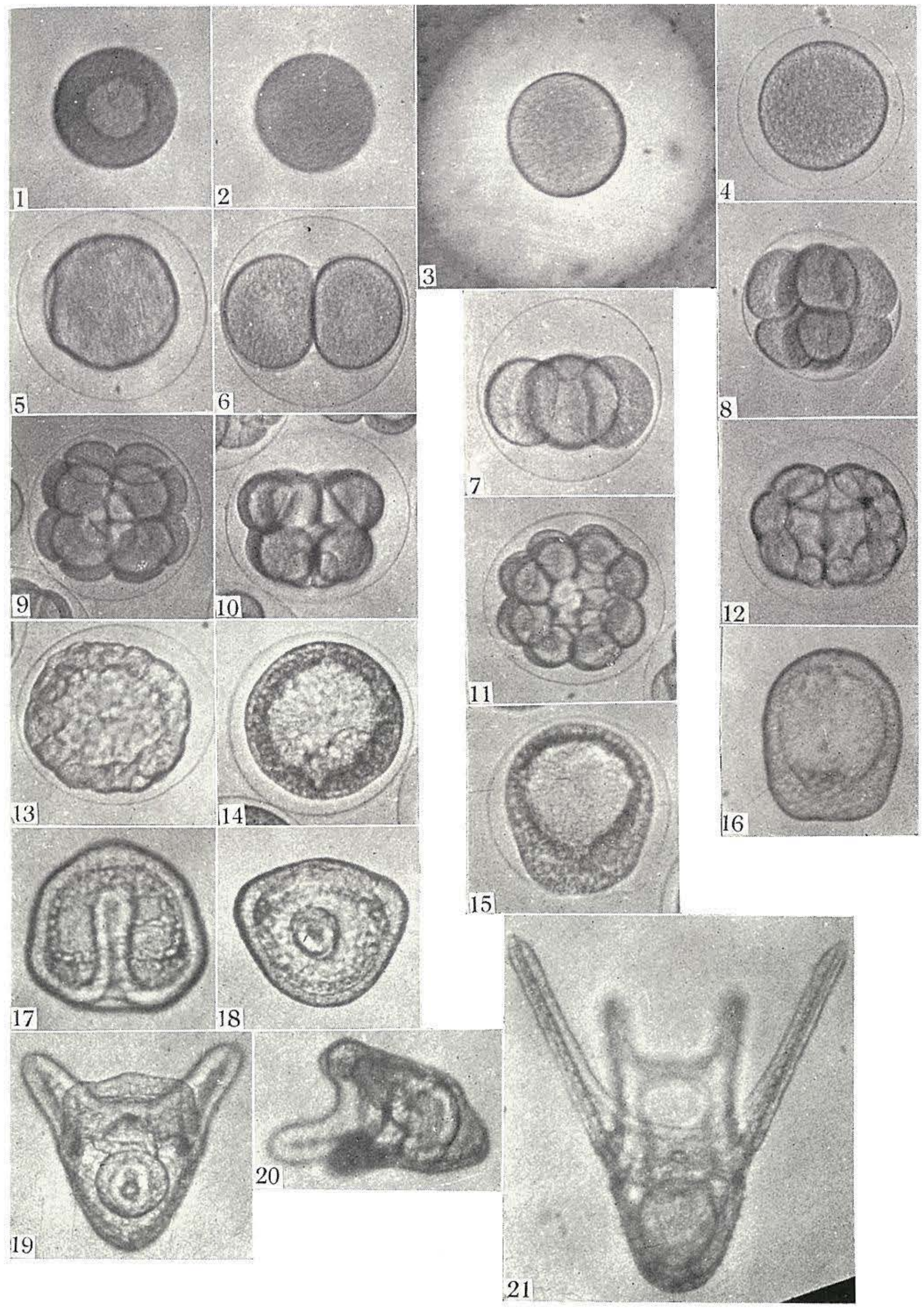

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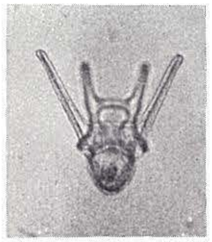
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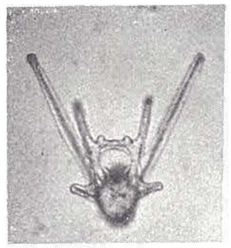
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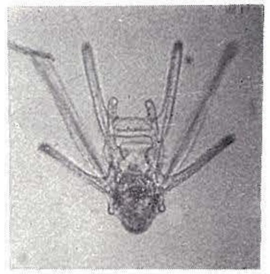
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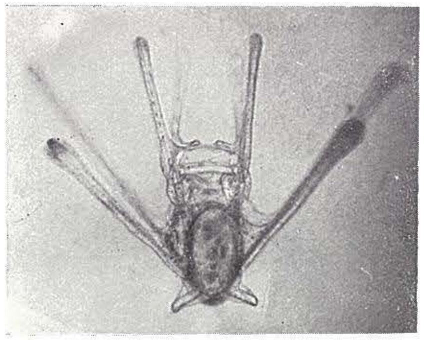
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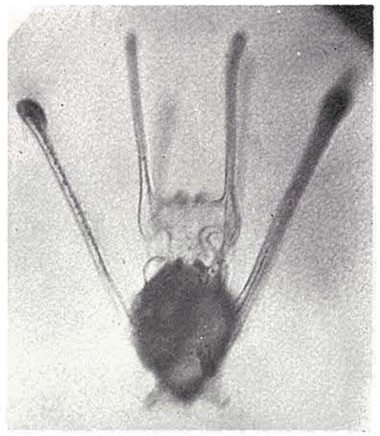
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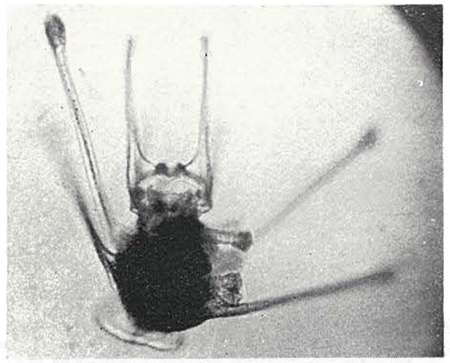
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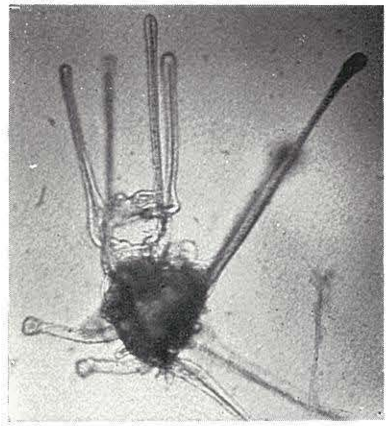
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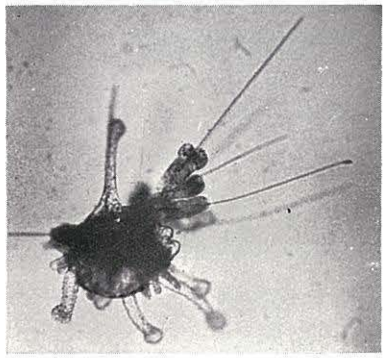

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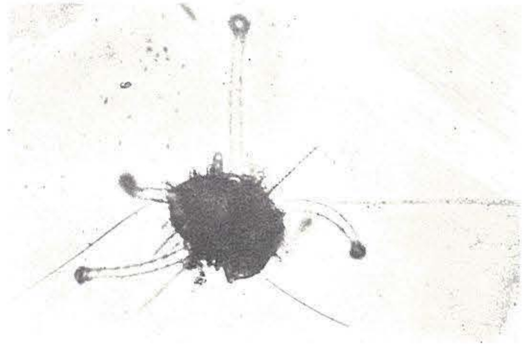

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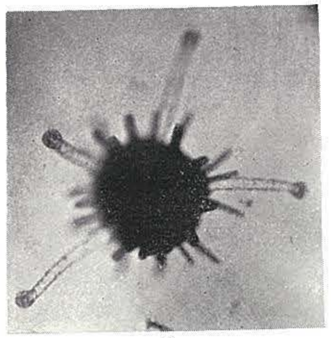

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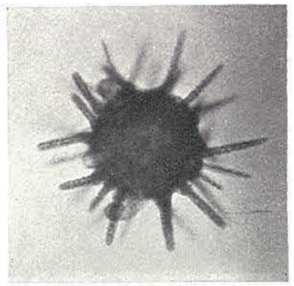

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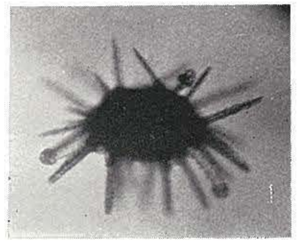

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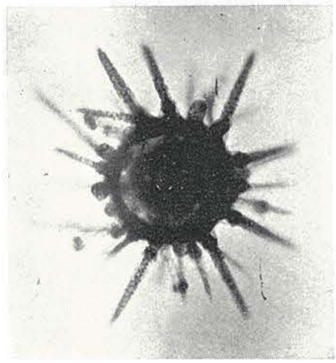

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